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Resveratrol: Mechanistic and Therapeutic Perspectives in Pulmonary Arterial Hypertension

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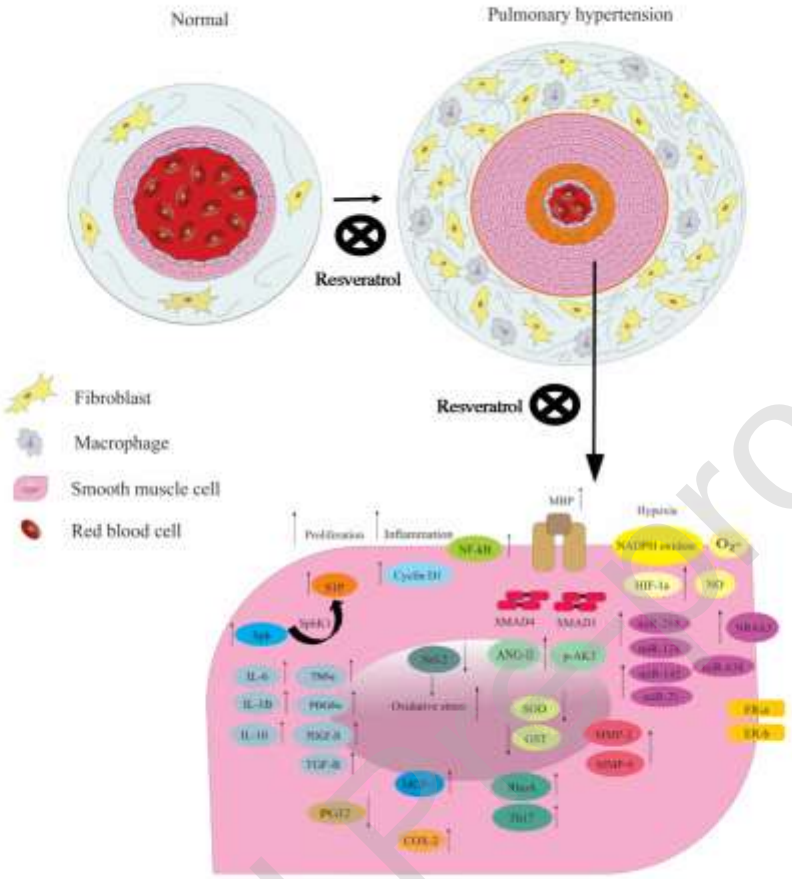
Graphical abstract



Abstract

Resveratrol, trans 3,5,4′-trihydroxystilbene, is a stilbenoid polyphenol with a wide range of properties including antioxidant, neuroprotective, cardioprotective, anti-inflammatory and anticancer activities. It is found in the skins of grape (50–100 μg/ml), red wine, peanuts, bilberries, blueberries and cranberries. The most important effects of resveratrol have been found in cardiovascular disease, with pulmonary arterial hypertension (PAH) being a major severe and progressive component. Many factors are involved in the pathogenesis of PAH, including enzymes, transcription factors, proteins, chemokines, cytokines, hypoxia, oxidative stress and

others. Resveratrol treats PAH through its actions on various signaling pathways. These signaling pathways are mainly suppressed SphK1-mediated NF-κB activation, BMP/SMAD signaling pathway, miR-638 and NR4A3/cyclin D1 pathway, SIRT1 pathway, Nrf-2, HIF-1 α expression, MAPK/ERK1 and PI3K/AKT pathways, and RhoA-ROCK signaling pathway. Resveratrol efficiently inhibits the proliferation of pulmonary arterial smooth muscle cells and right ventricular remodeling, which are underlying processes leading to enhanced PAH. While supportive evidence from randomized controlled trials is yet to be available, current *in vitro* and *in vivo* studies seem to be convincing and suggest a therapeutic promise for the use of resveratrol in PAH.

Keywords: resveratrol, pulmonary arterial hypertension, signaling pathway

Introduction

Phytoestrogens are naturally occurring compounds mostly found in soy, fruits and vegetables. This class of compounds contains bioactive molecules classified into four main groups, namely flavonoids, isoflavonoids, stilbenes and lignans. Stilbenes, in particular resveratrol, have been reported to be beneficial to human health by showing anticarcinogenic, antitumor, antioxidant and estrogenic/antiestrogenic activity [1]. Resveratrol was first isolated from white hellebore (*Veratrum grandiflorum O. Loes*) roots in 1940, then from *Polygonum cuspidatum* roots in 1963 and until now it has been detected in more than 70 plant species. In plants, resveratrol is synthesized in response to fungal attack, mechanical injury and ultraviolet (UV) irradiation. High

concentrations of resveratrol are present in the skins of grape in the range of 50–100 μg/ml due to fungal infection. It is also found in red wine, peanuts, bilberries, blueberries and cranberries [2].

Resveratrol, with the chemical (IUPAC name) of (trans 3,5,4′-trihydroxystilbene), is a stilbenoid polyphenol, possessing two phenol rings linked to each other by an ethylene bridge. Regarding chemical structure, resveratrol is identified in two isomeric forms, *trans* resveratrol and *cis* resveratrol, with the *trans* form isomerizing to the *cis* form when exposed to UV irradiation (Figure 1(a)). The trans form of resveratrol is dominant because of its greater prevalence and different biological activities. The nearly planar *trans* resveratrol has a rigid and less flexible conjugate network than *cis* resveratrol, which causes it to be stable under "accelerated stability" conditions of 40 °C and 75% humidity in the presence of air [3]. Three glycosylated forms of resveratrol have been isolated from plants, namely, piceid, resveratroloside and piceatannol glucoside (Figure 1(b,c,d)). These analogues were identified as being the major antibacterial compounds; they have similar antioxidant capacity as resveratrol, but show more powerful bioactivities. Glycosylated resveratrol analogues show biological effects as they can be hydrolyzed into deglycosylated forms after transepithelial passage. For example, piceid showed higher scavenging activity against hydroxyl radicals; however, resveratrol had a significant protective effect against H_2O_2-induced cell damage. It has been reported that piceatannol with one more hydroxyl group shows stronger anti-inflammatory, anti-proliferative, immunomodulatory, anti-leukemic, anti-leishmanial and protein-tyrosine kinase inhibitory effects. Resveratroloside is more effective against hepatitis B virus. [2, 4].

Resveratrol as is well known as a biologically active compound possessing a wide range of properties, including antioxidant, neuroprotective, cardioprotective, anti-inflammatory and anticancer actions [5-8]. Most importantly, resveratrol shows beneficial cardiovascular effects especially on pulmonary arterial hypertension (PAH) by ameliorating endothelial dysfunction and arteriolar remodeling [9, 10]. PAH is a severe and progressive disease caused by genetic defects in the bone protein signaling pathways, the most common cause of PAH is related to the inactivating mutations in the gene encoding bone morphogenetic protein type II receptor (BMPRII). BMPRII is a transmembrane serine/threonine kinase receptor that is vital for development, embryogenesis and adult tissue homeostasis. The pressure of the lungs in PAH patients exceeds 25 mm Hg at rest and 30 mm Hg with exercise. The main symptoms of this disease

are shortness of breath, fatigue, and chest pain [11, 12]. Single or multidrug therapies are applied to treat PAH such as anticoagulants, calcium channel blockers, diuretics, and prostanoids. Combination therapy is also regarded as a standard care in PAH [13, 14]. Smooth muscle cells (SMCs) have a key role in pathogenesis of PAH. SMCs de-differentiate, grow into the subendothelial space and secrete fibrous material causing intimal fibrosis (Figure 2A). However, many PAH patients still have poor prognosis and can experience systemic side effects such as liver damage, breathlessness, diarrhea, nausea, and pain [15]. Therefore, there is a need to identify safe and innovative therapeutics for PAH. This review introduces resveratrol as a new strategy to treat PAH and discusses approaches and mechanisms by which resveratrol may meet this aim.

1. Inhibition of pulmonary arterial remodeling

1.1 Suppression of SphK1-mediated NF-κB activation

The known mechanisms underlying PAH include persistent pulmonary vasoconstriction, thrombosis *in situ* and vascular remodeling. Current therapies have focused on vasoconstriction factors including NO inhalation, phosphodiesterase inhibitors, calcium channel blockers, prostanoids, endothelin receptor antagonists, all of which offer symptomatic relief. There is notable evidence suggesting that vascular remodeling, in which media hyperplasia and intima thickening occurs, plays a vital role in the pathogenesis of PAH. Abnormal proliferation of pulmonary arterial smooth muscle cells (PASMCs) is involved in this process [16, 17]. Sphingosine kinase 1 (Sphk1), an oncogenic key enzyme, is highly expressed in various types of human cancer and promotes tumor progression, proliferation, differentiation, invasion and metastasis [18]. SphK1 catalyzes the phosphorylation of sphingosine to generate sphingosine-1-phosphate (S1P), which is a bioactive lipid mediator. Abnormal activation of SphK1/S1P signaling has been known to be involved in PAH. Patients with idiopathic PAH possess elevated levels of SphK1 and S1P accompanied with PASMCs proliferation and pulmonary vascular structure remodeling (Figure 2B). Moreover, cyclin D1 expression has been found to be upregulated in PASMCs from monocrotaline (MCT) or hypoxia- induced PAH- model rats [19, 20]. Cyclin D1 is a protein which is encoded by the CCND1 gene and functions as a regulator of Cyclin-dependent kinase. During the G1 phase, Cyclin D1 is synthesized, accumulates in the nucleus and regulates

the G1/S phase transition and thus is responsible for cell proliferation [21]. Additionally, previous studies have demonstrated that Nuclear factor-kappaB (NF-κB), a transcription factor with a pivotal role in cell differentiation, proliferation and survival, is increased in PASMCs of PAH patients [22-24] (Figure 2B). The suggested mechanism in this regard showed that SphK1/S1P signaling increased NF-κB and promoted cyclin D1 expression in lung tissues, which then caused the induction of PASMCs proliferation, resulting in pulmonary vascular remodeling and development of MCT-induced PAH (Table 1). Resveratrol is able to suppress NF-κB activation, downregulate cyclin D, and inhibit MCT induced proliferation and migration of PASMCs (Figure 2B). A combination of resveratrol with PF543 (an SphK1 inhibitor) or with PDTC (NF-κB inhibitor) effectively reduced right ventricular systolic pressure (RVSP), diminished pulmonary vascular remodeling, and prevented the improvement of PAH [25].

1.2 Disruption of PDGF; normalization of BMP/SMAD signaling pathway

Structural changes (marked medial wall thickening) occurs in MCT-induced pulmonary hypertension contributing to pulmonary arterial resistance. MCT is a pyrrolizidine alkaloid extracted from the plant *Crotalaria spectabilis* to induce PAH in rats. Resveratrol was able to change the vessels to a normal morphology and reduce PASMCs proliferation without increment of apoptosis rate. The benefit of this to clinical studies of resveratrol is good tolerability and lack of adverse effects. Antiproliferative properties of resveratrol are due to the expression of heat shock protein HSP27, and tumor suppressor gene p53 in PASMCs [26]. In addition, it has been documented that MCT-induced PAH rats show increased ED-1 positive cells (a rat monocyte/ macrophage marker) in their lungs and considerable upregulation of growth factors and inflammatory cells, including IL-6, IL-1β, tumor necrosis factor-α, PDGF-α, PDGF-β, and TGF-β) (Figure 2C). Suppression of platelet derived growth factor (PDGF) signaling pathways prevents progression of MCT induced PAH (Table 1). Resveratrol treatment could effectively attenuate RVSP and pulmonary arterial remodeling by decreasing these cytokines and inflammatory cells (including PDGF-α/β) as well as ED-1–positive cells in the lung. However, the mechanisms by which resveratrol is involved in inflammatory process are not well understood. Importantly, recent studies have demonstrated that development of PAH is associated with expression and function of the bone morphogenetic protein (BMP)/SMAD signaling pathways [27]. BMP antagonists (chordin), BMP receptors (ACVR1 and ACVRL1) and SMAD signaling molecules (SMAD1/4)

antagonize PASMC proliferation. Resveratrol normalizes the alterations in the expression of many components of the BMP receptors. In addition, Csiszar *et al.* found that resveratrol could significantly improve endothelial dysfunction, attenuate oxidative stress and NADPH oxidase expression involved in small pulmonary arteries [24]. They suggested that downregulation of NADPH oxidase is related to debilitation of inflammatory cytokine production, as NADPH oxidase is regulated by inflammatory cytokines (tumor necrosis factor-α and TGFβ) [28, 29]. Moreover, upregulation by reseveretol of eNOS (endothelial NO synthase) (Figure 2C), which may also affect caveolin and/or BH4 levels and induction of heme oxygenase 1 in the pulmonary arteries of MCT-induced PAH rats was confirmed.

1.3 Downregulation of miR-638 and NR4A3/cyclin D1 pathway

Nuclear hormone receptors (NRs) have been identified as a superfamily specifically regulating metabolic function in cells and tissues. NR4A, important transcriptional regulators, belong to these family receptors; they control inflammatory processes in several diseases, including atherosclerosis and arthritis. The NR4A family comprises three kinds of mammalian nuclear receptors, including NR4A1 (Nur77), NR4A2 (Nurr1), and NR4A3 (Nor-1). However, these receptors have no known endogenous ligand(s) but can be activated by exogenous agents. Deficiency of NR4A2 and NR4A3 in murine and human myeloid cells provides proof of their function as enhancers of MIP-3α expression, while NR4A activity contributes to the suppression of lipopolysaccharide (LPS) induced MCP-1 gene and protein expression [30]. Previous studies have demonstrated that in vascular injury, NR4A3 plays a crucial role in neointima formation by promoting smooth muscle cell proliferation through regulating cyclin D1 [25, 31]. Additionally, many kinds of MicroRNAs (miRNAs) have been identified in pulmonary hypertension (miR-204, miR-124, miR-143, miR-21) and were shown to be involved in regulation of the NR4A superfamily (Figure 2C). miRNAs are classified as small (20–25nt) non-coding RNAs which have been identified to regulate a wide range of biological activities including differentiation, apoptosis and proliferation [32, 33]. Resveratrol regulates miRNA expression such as miR-193a, miR-34a, and miR-21 in allergic asthma, pulmonary fibrosis and acute lung injury. Due to the importance of miRNAs as post transcriptional regulators of gene transcription in pulmonary hypertension, including miR-124, miR-21, miR-143 and others, new strategies have emerged for pulmonary hypertension treatment. It has also been revealed that resveratrol has a crucial role in miR-638

expression in pulmonary vascular remodeling [34]. Expression of miR-638 consequently regulates gene expression pathways associated with autophagy, oxidative stress response, aging, and proliferation via down-regulating multiple genes such as SMC1A, CDK2, and DACT3 [35, 36]. Liu et al. introduced NR4A3 as a direct target of miR-638 in PASMCs and revealed that miR-638 is able to prevent proliferation through down regulation of NR4A3/cyclin D1 pathway (Figure 2C) [34]. Considering previous studies, they confirmed that NR4A3 is involved in both pulmonary vascular remodeling and systemic vascular disorders. However, the potential value of NR4A3 in PAH treatment remains uncertain table 1 [37, 38].

1.4 Regulation of Cell Cycle Regulators by Silence Information Regulator 1 (SIRT1)

PAH can occur in various conditions including a number of rheumatic diseases. Systemic lupus erythematosus (SLE), mixed connective tissue disease (MCTD) and systemic sclerosis (SSc) are associated with PAH. The low survival rate of PAH patients associated with rheumatic diseases emphasises the importance of early diagnosis and treatment. It has been shown that resveratrol is a silence information regulator 1 (SIRT1) activator which induces apoptosis in rheumatoid arthritis synovial cells (MH7A cells) in a SIRT1- dependent manner [39, 40]. SIRT1 is a nicotinamide adenosine dinucleotide (NAD) dependent histone deacetylase which performs a variety of functions in oxidative stress, including aging, tolerance and metabolism. SIRT1 removes acetyl groups from histones, coregulators and transcription factors [41]. Overexpression of SIRT1 inhibits vascular smooth muscle cell (VSMC) proliferation and migration and causes cell cycle arrest at G1/S transition [42]. Increased SIRT1 affects FOXO3-FOXM1 axis signaling pathways which are forkhead box transcription factors and crucial for differentiation, cell proliferation, cell survival, DNA damage repair, senescence and cell cycle control, all of which are important features of PAH[43]. FOXM1 stimulates proliferation of developing smooth muscle cells and promotes pulmonary vascular development. FOXM1is an essential factor for PASMC proliferation and its overexpression stimulates proliferation and evasion of cancer like cells from apoptosis in PAH [44]. SIRT1 is activated by resveratrol, which then increases FOXO3. FOXO3 directly inactivates FOXM1at the transcriptional level ,which leads to cell cycle arrest, apoptosis and inhibition of PASMCs proliferation [43]. Moreover, SIRT1 regulates the transcriptional activity of runt-related transcription factor 2 (RUNX2), which is transcription factor associated with osteoblast differentiation [45]. RUNX2 is up-regulated in lungs, activates HIF-1α activation, leads

to proliferation, resistance to apoptosis and PAH-PASMCs differentiation into osteoblast-like cells [46]. Resveratrol inhibits proliferation and differentiation of PAH-PASMCs through mediation of Sirt-1/Runx2signaling pathway [47]. The other factor related to SIRT1 is poly-ADP-ribose polymerase-1 (PARP1). SIRT1 acts as an inhibitor of PARP1. In PAH patients inflammation and oxidative stress increase DNA damage leading to increased expression of PARP1 [48]. Treatment with resveratrol as potent agonists of PARP1 through inhibition of SIRT1 attenuates inflammatory markers [49]. Zhou *et al.* indicated that SIRT1 is involved in resveratrol- mediated prevention of PAH. SIRT1 regulates expression of the cell cycle regulators cyclin D1, p21 and cyclin E, and could arrest human PASMCs (HPASMCs) in G0/G1 phase via platelet-derived growth factor BB (PDGF-BB) treatment. They suggested that resveratrol prevents pulmonary arterial remodeling through SIRT1-mediated regulation of cyclin D1 and p21 expression (Table 1, Figure 2D) [50]. Preservation of p21 as a cell cycle inhibitor is an essential factor in suppressing MCT-induced PAH in rats. P21 causes G1 arrest in cells contributing to inhibition of PASMCs proliferation via angiotensin converting enzyme inhibitors. Cyclin D1 ameliorates G1-to-S phase progression of cell cycle as a mitogenic signal sensor [51, 52]. Zhou *et al.* found that PDGF-BB increased cyclin D1 but decreased p21 expression in HPASMCs; however, resveratrol reverses this effect. Resveratrol maintained p21 expression and increased cyclin D1 expression level in the lungs of MCT- induced PAH rats. In the case of cyclin E, resveratrol hindered the increase of its expression by PDGF-BB treatment. However, cyclin E is not as important as p21 and cyclin D1 in pulmonary arterial remodeling [50]. SIRT1 function was also investigated in hypoxia- induced PAH rats, where resveratrol in combination with SRT1720 (as a selective SIRT1 activator), significantly inhibited PASMCs proliferation by inducing nuclear pyknosis and mitochondrial swelling, thereby contributing to PASMCs apoptosis and prevention of pulmonary vascular remodeling. Notably, resveratrol could not affect the level of SIRT1 protein but had significant anti-proliferation and pro-apoptosis effect in PASMCs (Table 1) [53].

1.5 Restoration of Nrf-2

Oxidative stress is defined as a state in which reactive oxygen species (ROS) are accumulated in the cellular environment, resulting in damage of DNA, RNA and proteins. In renal and vascular tissues inflammation accompanied by oxidative stress plays an important role in the pathogenesis of hypertension. A mechanism by which cells fight against oxidative stress is activating the

transcription factor NF-E2-related factor 2 (Nrf-2). To promote the overall survival of cells, Nrf-2 induces transcription of genes which are responsible for detoxifying the ROS and removing damaged proteins [54]. Hence, impaired activation of Nrf-2 leads to amplification or development of inflammation and oxidative stress makes it essential to seek strategies to restore the activity of Nrf-2. With respect to the effective cardiovascular benefits of resveratrol in counteracting pro-inflammatory cytokines, arterial remodeling and amelioration of endothelial dysfunction have been observed in animal studies [9, 10]. Moreover, it has been reported that resveratrol improves hypertension, ameliorates remodeling of the small arteries and prevents the development of contractile dysfunction and cardiac hypertrophy in spontaneously hypertensive rats (SHR) [10, 55]. Javkhedkar *et al.* indicated that renal tissue of untreated SHR shows interstitial immune cell infiltration, involved with oxidative stress in renal proximal tubular epithelial cells. The untreated SHR significantly showed a reduction of nuclear Nrf-2, which has a major role in the pathogenesis of inflammation and oxidative stress in the SHR kidney. However, treatment with resveratrol, restored the natural compound activator of Nrf-2 (Table 1), decreased oxidative stress in proximal tubular epithelial cells, reduced the number of interstitial angiotensin (ANG) II-positive cells and inflammatory cells in the kidney and ameliorated progression of hypertension in SHR. Reduction of oxidative stress is related to Nrf-2 activity restoration and expression of antioxidant enzymes [56]. Antioxidant enzymes such as superoxide dismutase (SOD) and glutathione-S-transferase (GST) play major roles in protecting cells in the aging process and against oxidative injury [57]. Long-term administration of resveratrol enhances SOD and GST levels in the SHR (Figure 2B).

1.6 Inhibition of HIF-1 α expression via suppressing the MAPK/ERK1 and PI3K/AKT pathways

A common model among the 5 types of PAH, hypoxic pulmonary hypertension (HPH) is characterized by vasoconstriction of distal arterioles which acutely causes persistent elevation of pulmonary arterial pressure contributing to pulmonary artery remodeling and right ventricular hypertrophy [58]. Chronic hypoxia could significantly elevate right ventricular systolic pressure (RVSP), vascular remodeling, infiltration of inflammatory cells, PASMCs proliferation and reactive oxygen species in rats. A key factor in vascular remodeling is attributed to PASMCs proliferation, which is suppressed by resveratrol [59]. Studies have demonstrated that both

classical steroid hormone receptors, ERα and ERβ, are expressed on PASMCs as well as pulmonary artery endothelial cells (PAMCs) [60, 61]. Resveratrol was shown to activate ERs in vascular endothelial cells (ECs), in particular ER-α, which is involved in the cardiovascular protective effects of resveratrol. In addition, increased levels of serum estrogen in both ovaries has been observed after treatment with resveratrol [62-64].However, Xu *et al.* showed that hypoxia-induced proliferation of PASMCs was inhibited by resveratrol in an ER independent manner. The discrepancy might be because of the different cell types and animal disease models used and the various biological effects of resveratrol [59]. Studies show thioredoxin-1 (Trx-1) and nuclear factor erythroid-2 related factor 2 (Nrf-2) are upregulated in resveratrol treatment. Nrf-2 is a transcription factor which regulates redox balance and protects cells against inflammatory and oxidative lesions. The balancing effects of Nrf-2 is exerted through the regulation of antioxidant proteins and detoxification enzyme expressions such as HO-1 and Trx-1. Moreover, Nrf-2/Trx-1 axis modulates the apoptosis-related signaling pathway via binding to relevant factors [65-67]. It was shown that hypoxia reduces the expression of both Trx-1 and Nrf-2 *in vitro* and *in vivo*, which can then be upregulated by resveratrol treatment. By reversal of the Nrf-2/Trx-1 axis ROS production was decreased in cultured PASMCs [59]. During the progression of HPH Hypoxia-inducible factors (HIFs), especially HIF-1α, play an important role in modulating downstream gene transcriptions. Under hypoxia condition, HIF-1α is increased due to its oxygen-independent protein synthesis and oxygen-dependent degradation. Besides hypoxia, ROS and NO also activate HIF-1α production [68, 69]. Resveratrol and HIF-1α inhibitor (KC7F2) prevented the development of HPH through inhibition of HIF-1α, in which proliferation of PASMCs was decreased (Table 1). Other pathways which have been observed to activate HIF-1 α in different disease models are PI3K/AKT and MAPK/ERK1 pathways, which are increased in hypoxia condition [70, 71]. Resveratrol treatment could decrease HIF-1α, through inhibition of both pathways. Applying PD98059 and LY294002 as ERK1/2 and PI3K/AKT inhibitors also lead to the inhibition of phosphorylated ERK and AKT expression [59]. Protein kinase B (AKT), which is activated by phosphoinositide 3‑kinase (PI3K) and implicated in tumorigenesis, is a member of the serine/threonine protein kinase family. p-AKT responds to various stimuli such as growth factors, stress and protein phosphatases. Activation of AKT is vital to prevent PASMCs apoptosis which may be induced by hypoxia [72-74]. Guan *et al.* showed that resveratrol could inhibit proliferation and migration of PASMCs through blocking the PI3K/AKT signaling pathway (Table 1) [75]. It

has been demonstrated that ANG II may increase the expression of p‑AKT, contributing to the formation of PAH. p-AKT consequently activates the mTOR/P70S6K signaling pathway which further increases the expression of p‑P70S6K, causing proliferation and migration of PASMCs [76-78]. Guan *et al.* indicated that resveratrol has the potential of regulating cell cycles, as expression levels of cyclin‑dependent kinase inhibitors (p21 and p27) were rescued from hypoxia after resveratrol treatment. Moreover, resveratrol inhibited protein expression levels of matrix metallopeptidases (MMPs), including MMP‑2 and MMP‑9 [75]. MMPs play important roles in proliferation and metastasis of PASMCs and also may increase their migration and proliferation via activation of the PI3K/AKT signaling pathway figure 2 [79].

1.7 Inhibition of RhoA-ROCK signaling pathway

Ras homolog family member A (RhoA) is a Protein Coding gene of the Rho-GTPases family found in eukaryotic cells. RhoA is activated by the microtubule-associated guanine nucleotide exchange factor (GEF)-H, when it is released from microtubules initiating RhoA/Rho kinase/myosin light chain signaling pathway which regulates cellular contractility [80, 81]. GEF-H1, as a RhoA specific guanine nucleotide exchange factor, is related to the actin cytoskeletal structure, and microtubules regulates activity of RhoA. GEF Rho-associated protein kinase (ROCK) is an effector of the small GTPase Rho, belonging to the AGC family of serine-threonine kinases. ROCK is involved in various cellular functions such as vascular smooth muscle cell adhesion, proliferation and actin cytoskeleton organization [82]. The RhoA-ROCK signaling pathway activation is upregulated by different factors such as ANG II, endothelin-1, hypoxia and platelet derived growth factor in pulmonary hypertension [83]. Previous studies have demonstrated that RhoA-ROCK signaling pathway is associated with hypoxia-induced pulmonary hypertension and its activity inhibition modulates pulmonary hypertension (PH) in animal models and in humans [84] [85]. Cytoskeletal rearrangements are regulated through RhoA-ROCK signaling pathway in which downstream substrates, including myosin phosphatase target subunit (MYPT1), are activated [86]. In addition, CD4+T cells such as Th17 cells, play an important role in hypoxic triggered diseases. Naïve T cells differentiate into Th17 cells under hypoxic conditions and play a major role in development of chronic hypoxia-induced PH. It has been found that resveratrol could hamper Th17 activity and reduce the severity of autoimmune diseases [87, 88]. Moreover, SR1001, as a Th17 inhibitor, inhibited the development of chronic hypoxia-induced PH [89]. Li

et al. showed that RhoA expression of small intrapulmonary arteries was significantly reduced by SR1001 in HPH rats (Table 1). SR1001 also decreased the expression of p-MYPT1 and as a result ameliorated pulmonary vascular remodeling (Figure 2C). SR1001 in the combination of resveratrol treatment by inhibition of Th17 cell differentiation prevented the development of HPH [90].

2. Prevention of pulmonary trunk remodeling not right ventricular hypertrophy

The MCT model is frequently used to induce PAH in rats and examine the effects of pulmonary hypertension on right ventricular (RV) structure, metabolism and function. One single injection of MCT causes endothelial injury, vascular remodeling, and finally increased pulmonary vascular resistance and RV hypertrophy [91]. In the PAH model induced by MCT, beneficial effects of resveratrol have been investigated including decreased expression of pro-inflammatory cytokines, reduction in systolic pressures and RV wall thickness, vasorelaxant, and vasculoprotective effects in pulmonary arteries [24, 92]. However, the effect of resveratrol in both heart and pulmonary trunk remained undetermined. While previous studies have shown beneficial effects of resveratrol on RV remolding [92-94], only modest effects of it have been observed on cardiac remodeling, such as decreasing the total heart surface area. In the case of the pulmonary trunk, resveratrol prevented development of its medial hypertrophy. Wilson *et al.* showed that thickness of the tunica media and pulmonary truck lumen area were increased by 44% and 43%, respectively, but no significant effect occurred on cardiac remodeling. Effects on the sirtuin1- dependent pathway and severity of PAH due to the concentration of MCT used may be the reasons for these observations [95]. SIRT1, a member of the sirtuin family of proteins, showed anti-hypertrophic, cardiovascular benefits and life extending properties. Low expression of sirtuin1 lead to decreased production of antioxidants, impaired angiogenesis, increased expression of pro-apoptotic molecules, ischemia reperfusion injury and diastolic dysfunction [96, 97]. Sever PAH was induced using 60 mg/kg MCT in rats which exhibited 2-fold higher ANG II levels in plasma compared to control rats. The benefits of resveratrol might decrease in a more advanced form of PAH. In this study plasma levels of C-reactive protein, endothelin-1, aldosterone and ANG II were not changed by resveratrol. Various doses of resveratrol are used in the literature based on the initial weight of the specimen, MCT dosages and the duration of the study. In a MCT- induced PAH rat model study, a 20

mg/kg/day dose of resveratrol did not show any significant differences between PAH and PAH + resveratrol groups in increased lung and heart weight, which means a higher dosage of resveratrol is required. Resveratrol had limited effect on lung histopathology, suggesting that improving the administration route and using higher doses of resveratrol could be an effective way to improve the results. Resveratrol not only hampered systolic failure but also enhanced RV function. It was found that PAH model is accompanied by changes; compromised RV function, isolated RV cardiomyocytes, and myocyte hypertrophy. Consistent with these changes, increased remodeling markers were observed, including BNP, Tnn1c, collagen 1, the anti-inflammatory cytokine IL-10 and the inflammatory IL-1β. Resveratrol treatment decreased the inflammatory markers and modified a decrease of the remodeling effect (Table 1). These results showed resveratrol mediated SIRT1 upregulation with a decrease in the acetylation profile [98].

3. Downregulating monocyte chemoattractant protein-1 (MCP-1)

Previous studies have highlighted vascular-related effects of resveratrol, including suppression of platelet aggregation [99], inhibition of low-density lipoprotein oxidation [100, 101], activation of endothelial nitric oxide synthase (eNOS) [102], inhibition of leukocyte adhesion [103], inhibition of tissue factor expression [104], and reduction in vascular cell adhesion molecule-1 expression [105]. Taken together, resveratrol potentially inhibits several key steps in the atherogenic process and have protective effect on cardiovascular disease. Pulmonary thromboembolism (PTE), defines a life-threatening condition associated with acute lung vascular injury and PAH. Recent studies have found that PTE would cause an inflammatory response in which cytokines and chemokines are produced by infiltrating inflammatory cells and could play major role in the formation of PAH. Among these chemokines and cytokines, monocyte chemoattractant protein-1 (MCP-1) has been shown to be the most effective inflammatory modifier of pulmonary inflammation and blood vessel which might be related to the formation of PAH. MCP-1 is significantly elevated during the early stages of PTE [106-108]. It is secreted by mononuclear cells, different non-leukocytic cells such as endothelial cells, vascular smooth muscle cells (VSMC) and resident renal cells. To find whether MCP-1 could be associated with the formation of acute PTE-induced PH, Chen *et al.* used a rat model of acute PTE and treated them with C1142, a rodent chimeric monoclonal antibody (mAb) that neutralizes MCP-1. Immunohistochemistry results showed that C1142 could decrease

the values of mean pulmonary artery pressure (mPAP). Resveratrol acts as an inhibitor for synthesis and secretion of MCP-1 in vascular endothelial cells. However, little is known how resveratrol downregulates MCP-1 and modulates PAH. Inhibition of p-p38MAPK activation has been found to reduce MCP-1 levels [109]. In the rat model of acute PTE, diminished acute PTE-induced p-p38MAPK and consequently MCP-1 expressions was significantly observed by co-incubation of SB203580 and resveratrol. SB203580 was utilized as a p-p38MAPK specific inhibitor which showed synergistic effect in combination with resveratrol (Table 1) [110].

4. Accumulation of heat shock protein 90 (HSP90) in PAH-PASMCs

HSP90 is a cytosolic chaperone involved in stability and folding of proteins. In stressful conditions which may lead to accumulation of damaged proteins, the HSP90 chaperone machinery is overexpressed in cancer cells, consequently protecting an array of oncoproteins from degradation and misfolding implicated in PAH progression and development. HSP90 is also compartmentalized in mitochondria rather than cytosol, but not in normal cells. It has been demonstrated that compartmentalized HSP90 has a crucial role in PAH, regulating the cancer like phenotype of PASMCs [34]. The effect of resveratrol on HSP90 has been investigated in some diseases, however no studies were found in the case of PAH. Resveratrol enhanced both renal function and survival in a rat model of uremia, associated with increased expression of Hsp70, Hsp90, Hsp27, Hsp25, Hsp40 and Hsp60 [111]. Resveratrol also alleviated the jejunum mucosa injuries and improved intestinal morphology via modulating mRNA and protein expression of HSPs (HSP70, HSP90), epidermal growth factor and NF-κB [112]. It has been shown that resveratrol could enhance the apoptosis induction in chronic myelogenous leukemia through downregulation of HSP70 levels [113]. Although these studies show the effect of resveratrol on HSPs, no investigation has been conducted on its effect in PAH animal models.

5. 3,4′,5-trans-trimethoxystilbene (TMS)

In recent studies into PAH treatment, there has been increased interest in using resveratrol due to its various activities, including cell cycle blockade, estrogen like effect, antioxidant effects, inhibitory effects against AP-1, nuclear factor kappa B (NF-κB), matrix metalloproteinases

(MMPs), tumor necrosis factor- (TNF-) α, cyclooxygenase- (COX-) 2, and interleukin-(IL-) 1β activities. However, these beneficial effects are limited because of its poor pharmacokinetic profile, including its short half life in plasma (11.5 min), being poorly absorbed and easily oxidized [114]. To overcome these limitations, natural analogues of resveratrol, mainly TMS, have emerged as strong candidates (Figure 1,e). Moreover, TMS inhibits TNF-a-induced PASMC proliferation more potently than resveratrol. In comparison to resveratrol, TMS shows an enhanced anticancer profile, including improved induction of cell cycle arrest, cancer cell proliferation inhibition, increased apoptosis and reduced angiogenesis [115]. Moreover, it has been found that TMS possesses higher lipotropy and extended half-life, which makes it a better candidate for systemic applications compared with resveratrol. TMS and other resveratrol derivatives could occur naturally or be synthesized by resveratrol methylation (Table 1) [116]. Here we investigate the effect of TMS on PASMCs and also the mechanisms involved in improvement of PAH. Since PASMCs are the main targets for the treatment of PAH, several studies investigated the effect of resveratrol and TMS on PASMC proliferation. The results showed the potent inhibition effect of TMS on PASMC proliferation, which is induced by cell apoptosis. It was also demonstrated that TMS is a more effective anti-inflammatory compound than resveratrol to treat PAH. Induction of apoptosis for TMS was dose-dependent, with a potency 10 times higher than resveratrol [116]. Furthermore, it has been also found that the expression level of NF-κB, TNF-α, and IL-6 in peripheral blood serum, myocardial protein, and myocardial cells of the TMS-treated rats, was lower than untreated groups. NF-κB produces chemical factors, cytokines, immune receptors and cell adhesion molecules. Additionally, the expression of vasoactive substances (eg, PGI_2) is inhibited by NF-κB. TNF-α decreases the synthesis of prostaglandins (PGs) in PASMCs, increases pulmonary vascular reactivity, and induces pulmonary vasoconstriction. With the occurrence of PAH, the level of COX-2, the primary step in the synthesis of PG, increases, IL-6 is induced by TNF-α and IL-1β in mononuclear phagocytes when tissue is damaged. TMS successfully could inhibit these factors in peripheral blood and myocardial tissue as a potential new strategy for PAH treatment (Table 1) [117].

5.1 Inhibition of NOX/VPO1 Pathway

It is well established that most of drugs for PAH treatment including prostacyclin analogs, calcium channel blockers and endothelin receptor inhibitors improve vascular diastole function by targeting pulmonary artery and mainly provide symptomatic relief. As a novel strategy to treat PAH, there has been growing interest in anti-oxidant therapy. Oxidative stress, a steady state in which excess synthesis of reactive oxygen species (ROS) causes damage to cells or tissues, contributes to the development of PAH [118]. ROS, including radical or non-radical species ($O_2^{\cdot -}$, $OH^{\cdot}$, H_2O_2, and HOCl) are generated from multiple metabolic and enzymatic sources. Two most important enzymes, vascular peroxidase1 (VPO1), and NADPH oxidase (NOX), have been reported to play major roles in promoting oxidative stress. The NOX/VPO1 pathway is involved in angiotensin II-induced smooth muscle cell proliferation, myocardial ischemia/reperfusion injury, and hypertensive vascular remodeling [119-121]. Resveratrol in one study reduced right ventricular systolic pressure (RVSP) and pulmonary arterial remodeling, which was related to the downregulation of NOX1 and NOX2 expression [24] (Figure 2C). TMS, as a novel analog of resveratrol, also downregulated the expression of VPO1, NOX2, and NOX4 in pulmonary artery or RV and decreased their levels in plasma and lung tissue. TMS caused a decrease in hypoxia-induced H_2O_2 production which can be further catalyzed by VPO1 to form HOCl. It has been demonstrated that NOX-derived H_2O_2 consequently induces VPO1 protein expression via JNK/p38 MAPK-dependent signaling pathways [119]. TMS could significantly attenuate hypoxia-induced PVR and RV hypertrophy at both dosages (5 or 10 mg/kg). The therapeutic effect of TMS at a dosage of 10 mg/kg was similar to that of a dosage of 25 mg/kg for resveratrol. The results proved that TMS is a more potent therapy for PAH, as resveratrol did not show any significant effect at the dosage of 10 mg/kg table 1[122].

6. Inhalable resveratrol microparticles

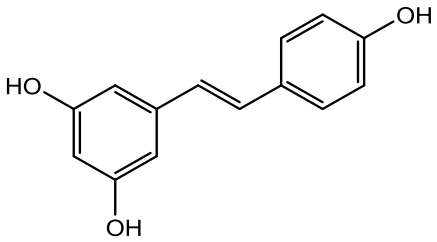
Conventional treatment approaches for PAH mainly utilize intravenous and oral routes, potentially leading to major systemic side effects such as liver damage, pain, diarrhea, nausea, and breathlessness. Interestingly, the inhalation route could be an alternative to increase bioavailability compared with oral administration. Additionally, it is more patient friendly due to its painless and non-invasive nature compared to the intravenous route [15, 123]. To control drug release and improve the treatment a potential approach is using polymeric carriers such as microparticles.

Buchi Labortechnik developed a technique by which dry powders with aerodynamic diameter less than 5 µm could be efficiently deposited in the lower respiratory tract. The Nano Spray Dryer B-90® has been introduced as a new generation of spray dryers which produces powders with reduced particle sizes and better yields [124, 125]. A Nano Spray Dryer was used to deliver resveratrol to the deepest lung areas, with the strategy of applying polymeric microparticles aimed at a controlled release formulation. For this purpose, a poly(ε-caprolactone) polymer was chosen as a water-insoluble shell structure with a molecular weight of 42.5 kDa to control the drug release rate. Trehalose and sodium deoxycholate were utilized as drying adjuvant and surfactant, respectively. Sodium deoxycholate was used to avoid particle agglomeration and increase the redispersion of the microparticles and flowability. The piezoelectric atomization technique was used to obtain particles with aerodynamic diameters of 2.32 µm and fine particle fraction of 50%. This process provided preferential yields (80%), with the powder moisture less than 2.0%. Aerodynamic properties of microparticles showed that they are suitable for drug deposition on the alveolar regions of lungs due to the increased flowability and low density of the powders. *In vitro* sustained release profiles proved the potential of inhaled administration of resveratrol for treatment of PAH table 1 [126].

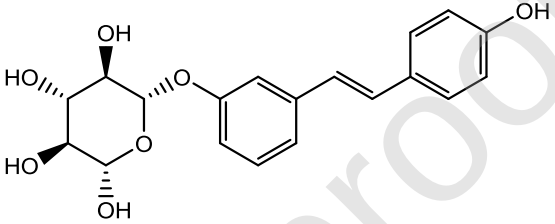
Conclusion

Pulmonary arterial hypertension is a progressive disorder characterized by high blood pressure in the arteries of lungs. The most common treatments for PAH over the past 15 years interfere with the endothelin, nitric oxide and prostacyclin pathways. Endothelin receptor antagonists which inhibit vasoconstriction and proliferation of PASMCs consist of bosentan, ambrisentan and macitentan. The nitric oxide pathway is divided into two categories; phosphodiesterase 5 inhibitors (sildenafil, tadalafil) and soluble guanylate cyclase stimulators (riociguat). Finally, in the prostacyclin pathway prostacyclin analogues (epoprostenol, treprostinil, iloprost) and the non-prostanoid IP receptor agonist (selexipag) have been used. Although these treatments improve quality of life and survival in patients, they do not cure the disease and an approach to improve the RV function remains unsolved. Resveratrol, a polyphenolic compound with anti-fibrotic and anti-inflammatory effects, has received considerable attention due to its protective properties in several

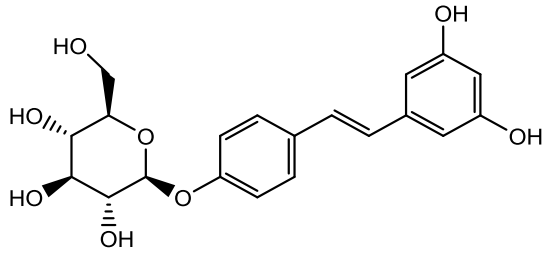
cardiovascular diseases. In this review we discuss recent research into the effects and mechanisms of resveratrol on PAH. Resveratrol shows anti-proliferative effects by inhibition of PASMCs proliferation leading to vascular remodeling through various signaling pathways in which many enzymes, proteins and transcription factors are involved. SphK1, S1P, cyclin D1, NF-κB, PAKT, PI3K are increased in PASMCs, and all are attenuated by resveratrol. Accumulating evidence shows that resveratrol decreases chemokine and cytokine driven inflammatory processes induced by mediators TGF-β, PDGF-α, PDGF-β, IL-1β, IL-18, IL-8, IL-10, and particularly IL-6, contributor to the development of PAH. In hypoxia conditions, especially chronic hypoxia, pulmonary arteries undergo vasoconstriction and then vascular remodeling characterized by intimal thickening. Resveratrol effectively reduced the activation of HIF-1α and HIF-2α as well as over-activation of mTOR in hypoxia conditions which contribute to the PASMCs differentiation and proliferation. Furthermore, resveratrol showed its potential to normalize miRNAs such as miR-124, miR-21, miR-143, which have been reported to be dysregulated in patients with PAH. The results of studies reviewed in this paper support the benefit of resveratrol as a new strategy for treatment of PAH.



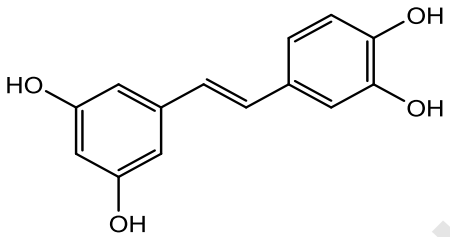
a) trans 3,5,4′-trihydroxystilbene (resveratrol)



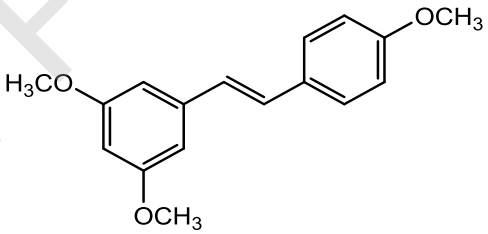
b) piceid



c) resveratroloside



d) piceatannol



e) 3,4′,5-trans-trimethoxystilbene (TMS)

Figure 1. Chemical structure of resveratrol and its derivatives: (a) resveratrol, (b) piceid, (c) resveratroloside, (d) piceatannol, (e) TMS

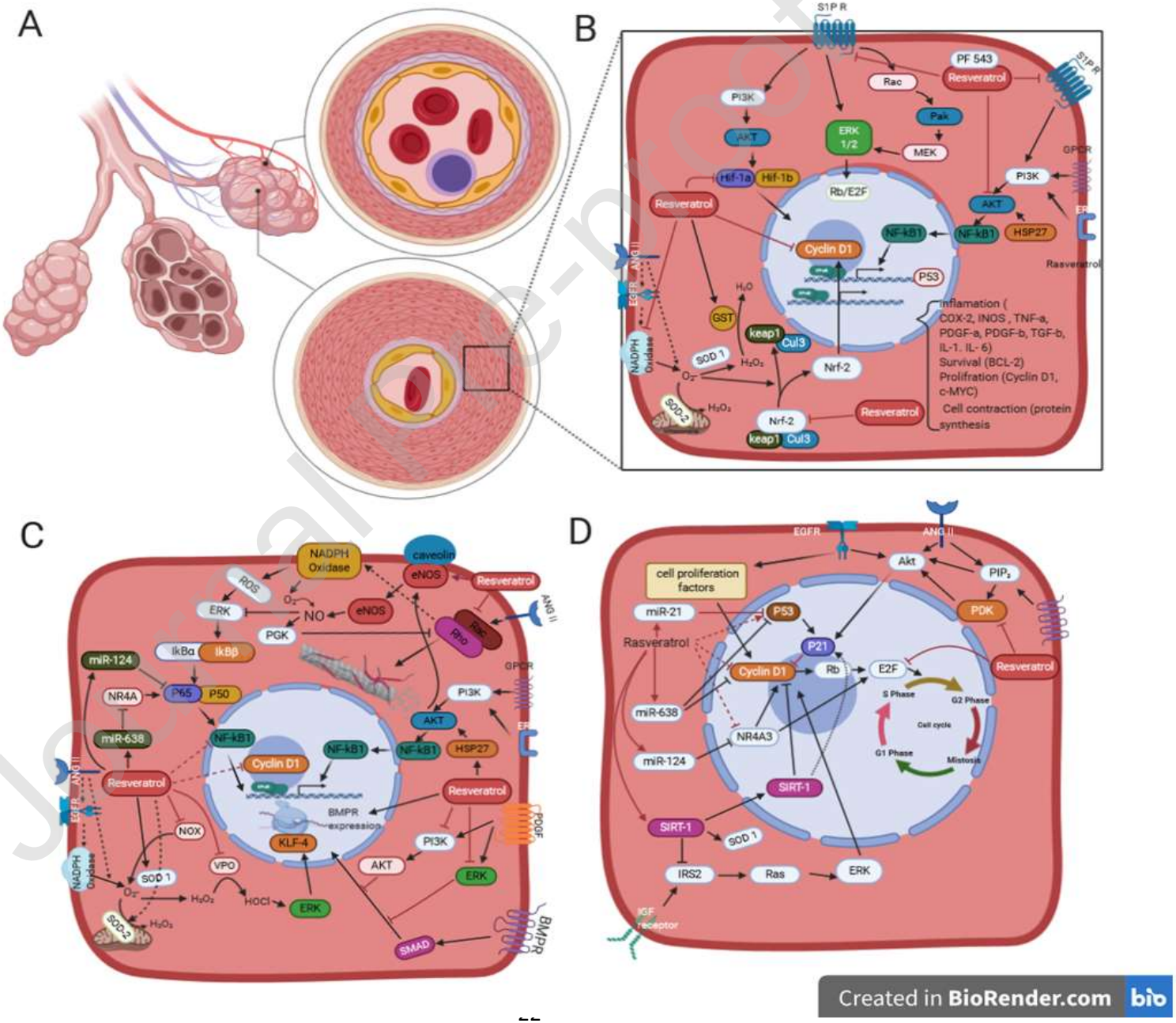


Figure 2. The figure shows signaling pathways involved in proliferation of PASMCs, and the effect of resveratrol on these pathways. Resveratrol inhibits SphK1, NF-kB signaling pathways (Figure 2B). Resveratrol induces BMPR expression in PASMCs which activates SMAD molecules downstream of the PMPR signaling pathway, leading to induction of anti-proliferative signals in these cells. Resveratrol also inhibits ERK and PI3K activities downstream of PDGFR signaling pathway which in turn reduces the proliferative effect of PDGF on pulmonary perivascular muscle cells. Downregulation of ERK activities reduce the inhibition effect of ERK on NO synthesis by eNOS enzyme. eNOS contains MAPK binding site which can control eNOS activities. Phosphorylation of eNOS by ERK reduces NO synthesis in muscle cells (Figure 2C). Resveratrol reduces PASMCs proliferation through enhancing miR638 and miR124 expression. These micro RNAs interfere with P65 expression downstream of NR4A signaling pathway that activate NF-kB molecule in PASMCs. As a result, NR4A signaling pathway is downregulated in PASMCs (Figure 2C). Resveratrol enhances SIRT-1 activity, leading to suppression of cyclin-D1 and activation of P21 function. SIRT-1, also enhances SOD function, which reduces ROS and pro-inflammatory effects on cell proliferation (Figure 2D). In oxidative stress and hypoxic conditions, ROS accumulates in cytoplasm, contributing to the increase of cell proliferation activity. Resveratrol enhances Nrf-2 activity. Resveratrol reduces ROS through enhancing Nrf-2 activity which increases antioxidant enzymes (SOD, GST) (Figure 2B). Stimulation of Rho/Rack signaling pathway with stimulatory receptors such as ANG II, endothelin-1, hypoxia, PDGF and Th17, provoke cytoskeleton rearrangement in pulmonary vascular muscle cells. Resveratrol downregulates expression of p-MYPT1 downstream of Rho/Rack signaling pathway and as a result ameliorates pulmonary vascular remodeling (Figure 2C). NOX/VPO-1 enzymes produce ROS that induce MAPK dependent signaling pathway, leading to PASMCs proliferation. Resveratrol and its analog TMS attenuate MAPK signaling pathway through reducing H_2O_2 production (Figure 2C).

Table 1. The effects of resveratrol on PAH: signaling pathways, dosage and route of administration.

Mechanism	Dose of treatment	Route of administration	Method of PAH induction	Outcome	Ref
Suppression of SphK1-mediated NF-κB activation	Resveratrol (25 mg/kg/day) PF543 (0.7 mg/kg/day) PDTC (100 mg/kg/day)	i.p	PAH was induced by a single intraperitoneal injection of MCT (60 mg/kg) on day 1 into male SD rats	PAH was mediated through inhibition of the SphK1/S1P/NF-κB/cyclin D1 signaling pathway	[25]
Disruption of PDGF, normalization of BMP/SMAD signaling pathways	Resveratrol (25 mg/kg/day)	PO	PAH was induced by MCT (60 mg/kg SC) in male SD rats	-Suppression of PDGF, and the BMP/SMAD signaling pathways -Decreasing ED-1–positive cells -Attenuation of NADPH oxidase -Upregulation of eNOS	[24]
Downregulation of miR-638 Up-regulation of NR4A3	Resveratrol (25 mg/kg/day)	PO	PAH was induced by a single subcutaneous injection of 60 mg/kg of MCT into adult male Wistar rats	-Prevention of MCT-induced pulmonary vascular remodeling - Suppression of PASMCs proliferation through miR-638 up-regulation	[34]
Regulation of cell cycle regulators (cyclin D1, p21) by SIRT1	Resveratrol (2.5 mg/kg/day and 20 mg/kg/day)	PO	Adult male SD rats received a SC injection of MCT at a dose of 60 mg/kg	- Attenuation of pulmonary arterial remodeling - Decreased pulmonary arterial pressure - Upregulation of SIRT1 and p21 expression - Downregulated cyclin D1 expression	[50]
Activation of SIRT1	Resveratrol (25 mg/kg/day) SRT1720 (25 mg/kg/day)	-Resveratrol was administered by gavage - PASMCs were exposed to SRT1720 for 24-48h	Male Wistar rats inspired oxygen of Fio_2: 0.21 and Fio2: 0.12 for 21 days	-Activation of SIRT1 and inhibition of PASMC proliferation -Improving RVSP - Reversing pulmonary arterial remodeling	[53]
Restoration of Nrf-2	Resveratrol (50 mg/l)	PO	Spontaneously hypertensive rats (SHR) administered resveratrol from 3–4 wk until 12 wk of age	-Reduction of ANG II-positive cells and inflammatory cells in kidney - Restoration of Nrf-2 activity - Expression of SOD and GST	[56]

Inhibition of HIF-1 α	Resveratrol (40 mg/kg/day)	Gavage administration	SD rats were exposed to hypoxia for 28 days along with resveratrol treatment	-Alleviation of right ventricular systolic pressure and pulmonary arterial remodeling - Suppression of inflammatory cell infiltration - Decreased ROS production, suppressed tumor necrosis factor α, interleukin 6, and interleukin 1β - Suppression of the MAPK/ERK1 and PI3K/AKT pathways, inhibition of the HIF-1 α expression	[59]
Inhibition of RhoA-ROCK signaling pathway	Resveratrol (40 mg/kg/day) SR1001 (25 mg/kg/day)	i.p	Adult male SD rats were exposed to a concentration of (10 ± 0.5)% O_2 for 21 days	- Inhibition of Th17 activity - Alleviated mean PAP, right ventricular hypertrophy, and hypoxic pulmonary vascular remodeling -Combination therapy alleviated HPH	[90]
Inhibition of PI3K/AKT signaling pathway	Resveratrol (10, 30 and 100 μmol/l)	Resveratrol was added to culture media of PASMCs	Hypoxic environment for PAMSCs was induced using an autonomous plexiglass chamber supplied with 5% CO2 and 95% N2 at 20 ml/min.	-Inhibition of hypoxia‐induced proliferation and migration of PASMCs - Inhibition of activation of the PI3K/AKT signaling pathway - Inhibition of MMP‐2 and MMP‐9	[75]
Effects on the sirtuin1 dependent pathway	Resveratrol (25 mg/kg/day)	PO	PAH was induced by MCT (60 mg/kg SC) in male SD rats	-Increased total heart surface area and RV lumen area -No significant effect on cardiac remodeling	[97]
Prevention of right ventricle remodeling	Resveratrol (20 mg/kg/day)	gavage administration	PAH was induced by a single subcutaneous injection of MCT (60 mg/kg body weight) into male SD rats	- protective effect against pathological remodeling changes and ventricular dysfunction -Improved right ventricular remodeling and function -Restoration of right ventricular cardiomyocyte structure and contractile function	[98]
Downregulation of MCP-1 by inhibition of p-p38MAPK	Resveratrol (10 mg/kg/day) SB203580 (2.5 mg/kg/day) C1142 (2 mg/kg/day)	i.p	Vascular obstruction by making thrombus in rats	- Downregulation of of MCP-1 and p-p38MAPK protein expression	[110]

				was shown by immunohistochemistry - decreased MCP-1 mRNA expression was revealed by Real-time PCR analysis	
Inhibition of PASMC proliferation by induction of apoptosis	TMS (20 μM) TNF-α (100 pg/ml)	TMS was added to cultured media of PASMCs	PASMCs were isolated from pulmonary arteries of SD male rats and cultured with exposure to TMS	Potent inhibitory effect of TMS on PASMC proliferation	[116]
Inhibition of NF-κB, TNF-α, and IL-1β	TMS 120 mg/(kg/day)	PO	PAH was induced by MCT (30 mg/kg SC) in male and female SD rats	Reduction of inflammatory factor levels in pulmonary hypertensive rats	[117]
Inhibition of NOX/VPO1 Pathway	TMS (5 or 10 mg/kg)	i.g	Rats continuously exposed to hypoxia (10% O_2) for 4 weeks	-Downregulation of VPO1, NOX2, and NOX4 expression -Attenuation of hypoxia-induced pulmonary vascular remodeling and RV hypertrophy	[122]
Atomization spray drying	Resveratrol (0.1 g)	Inhalation	-	Controlled release of resveratrol	[126]

MCP-1: monocyte chemoattractant protein-1, p-p38MAPK: p38 mitogen activated protein kinase, SB203580: p38MAPK specific inhibitor, C1142: rodent chimeric mAb, MCT: monocrotaline, i.p: intraperitoneal injection, PO: Per os (orally), SC: subcutaneous, PDGF: platelet derived growth factor, BMP: bone morphogenetic protein receptor, eNOS: endothelial NO synthase, SIRT1: silence information regulator1, RVSP: right ventricular systolic pressure, ANG: angiotensin, SOD: superoxide dismutase, GST: glutathione-S-transferase, PAP: pulmonary artery pressure, HPH: Hypoxic pulmonary hypertension, TMS: 3,4′,5-trans-trimethoxystilbene, i.g: intragastric, SD: Sprague Dawley rats, FIO_2: Fraction of inspired oxygen

Conflict of interests: None

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References

[1] H. Kalantari, D.K. Das, Physiological effects of resveratrol, Biofactors 36(5) (2010) 401-406.
[2] B. Salehi, A.P. Mishra, M. Nigam, B. Sener, M. Kilic, M. Sharifi-Rad, P.V.T. Fokou, N. Martins, J. Sharifi-Rad, Resveratrol: A double-edged sword in health benefits, Biomedicines 6(3) (2018) 91.
[3] M.H. Keylor, B.S. Matsuura, C.R. Stephenson, Chemistry and biology of resveratrol-derived natural products, Chemical reviews 115(17) (2015) 8976-9027.
[4] M. Li, K.R. Kildegaard, Y. Chen, A. Rodriguez, I. Borodina, J. Nielsen, De novo production of resveratrol from glucose or ethanol by engineered Saccharomyces cerevisiae, Metabolic Engineering 32 (2015) 1-11.
[5] D. Su, Y. Cheng, M. Liu, D. Liu, H. Cui, B. Zhang, S. Zhou, T. Yang, Q. Mei, Comparision of piceid and resveratrol in antioxidation and antiproliferation activities in vitro, PloS one 8(1) (2013).
[6] S. Fabris, F. Momo, G. Ravagnan, R. Stevanato, Antioxidant properties of resveratrol and piceid on lipid peroxidation in micelles and monolamellar liposomes, Biophysical chemistry 135(1-3) (2008) 76-83.
[7] A.M. Rimando, N. Suh, Biological/chemopreventive activity of stilbenes and their effect on colon cancer, Planta medica 74(13) (2008) 1635-1643.
[8] L. Kuršvietienė, I. Stanevičienė, A. Mongirdienė, J. Bernatonienė, Multiplicity of effects and health benefits of resveratrol, Medicina 52(3) (2016) 148-155.
[9] J. Behbahani, S.J. Thandapilly, X.L. Louis, Y. Huang, Z. Shao, M.A. Kopilas, P. Wojciechowski, T. Netticadan, H.D. Anderson, Resveratrol and small artery compliance and remodeling in the spontaneously hypertensive rat, American journal of hypertension 23(12) (2010) 1273-1278.
[10] S.J. Thandapilly, P. Wojciechowski, J. Behbahani, X.L. Louis, L. Yu, D. Juric, M.A. Kopilas, H.D. Anderson, T. Netticadan, Resveratrol prevents the development of pathological cardiac hypertrophy and contractile dysfunction in the SHR without lowering blood pressure, American journal of hypertension 23(2) (2010) 192-196.
[11] M.M. Hoeper, H.J. Bogaard, R. Condliffe, R. Frantz, D. Khanna, M. Kurzyna, D. Langleben, A. Manes, T. Satoh, F. Torres, Definitions and diagnosis of pulmonary hypertension, Journal of the American College of Cardiology 62(25 Supplement) (2013) D42-D50.
[12] S. Perrin, M.-C. Chaumais, C. O'Connell, D. Amar, L. Savale, X. Jaïs, D. Montani, M. Humbert, G. Simonneau, O. Sitbon, New pharmacotherapy options for pulmonary arterial hypertension, Expert opinion on pharmacotherapy 16(14) (2015) 2113-2131.
[13] N. Galie, A. Torbicki, R. Barst, P. Dartevelle, S. Haworth, T. Higenbottam, H. Olschewski, A. Peacock, G. Pietra, L.J. Rubin, Guidelines on diagnosis and treatment of pulmonary arterial hypertension: The Task Force on Diagnosis and Treatment of Pulmonary Arterial Hypertension of the European Society of Cardiology, European heart journal 25(24) (2004) 2243-2278.
[14] M. Burks, S. Stickel, N. Galie, Pulmonary arterial hypertension: combination therapy in practice, American Journal of Cardiovascular Drugs 18(4) (2018) 249-257.
[15] A. Saigal, W.K. Ng, R.B. Tan, S.Y. Chan, Development of controlled release inhalable polymeric microspheres for treatment of pulmonary hypertension, International journal of pharmaceutics 450(1-2) (2013) 114-122.

[16] M. Humbert, N.W. Morrell, S.L. Archer, K.R. Stenmark, M.R. MacLean, I.M. Lang, B.W. Christman, E.K. Weir, O. Eickelberg, N.F. Voelkel, Cellular and molecular pathobiology of pulmonary arterial hypertension, Journal of the American College of Cardiology 43(12 Supplement) (2004) S13-S24.
[17] K.R. Stenmark, K.A. Fagan, M.G. Frid, Hypoxia-induced pulmonary vascular remodeling: cellular and molecular mechanisms, Circulation research 99(7) (2006) 675-691.
[18] W. Li, C.-P. Yu, J.-t. Xia, L. Zhang, G.-X. Weng, H.-q. Zheng, Q.-l. Kong, L.-j. Hu, M.-S. Zeng, Y.-x. Zeng, Sphingosine kinase 1 is associated with gastric cancer progression and poor survival of patients, Clinical Cancer Research 15(4) (2009) 1393-1399.
[19] J. Chen, H. Tang, J.R. Sysol, L. Moreno-Vinasco, K.M. Shioura, T. Chen, I. Gorshkova, L. Wang, L.S. Huang, P.V. Usatyuk, The sphingosine kinase 1/sphingosine-1-phosphate pathway in pulmonary arterial hypertension, American journal of respiratory and critical care medicine 190(9) (2014) 1032-1043.
[20] J.R. Sysol, V. Natarajan, R.F. Machado, PDGF induces SphK1 expression via Egr-1 to promote pulmonary artery smooth muscle cell proliferation, American Journal of Physiology-Cell Physiology 310(11) (2016) C983-C992.
[21] H. Goto, A. Inoko, M. Inagaki, Cell cycle progression by the repression of primary cilia formation in proliferating cells, Cellular and molecular life sciences 70(20) (2013) 3893-3905.
[22] S. Hosokawa, G. Haraguchi, A. Sasaki, H. Arai, S. Muto, A. Itai, S. Doi, S. Mizutani, M. Isobe, Pathophysiological roles of nuclear factor kappaB (NF-kB) in pulmonary arterial hypertension: effects of synthetic selective NF-kB inhibitor IMD-0354, Cardiovascular research 99(1) (2013) 35-43.
[23] L. Li, C. Wei, I.-K. Kim, Y. Janssen-Heininger, S. Gupta, Inhibition of nuclear factor-κB in the lungs prevents monocrotaline-induced pulmonary hypertension in mice, Hypertension 63(6) (2014) 1260-1269.
[24] A. Csiszar, N. Labinskyy, S. Olson, J.T. Pinto, S. Gupte, J.M. Wu, F. Hu, P. Ballabh, A. Podlutsky, G. Losonczy, Resveratrol prevents monocrotaline-induced pulmonary hypertension in rats, Hypertension 54(3) (2009) 668-675.
[25] W. Shi, C. Zhai, W. Feng, J. Wang, Y. Zhu, S. Li, Q. Wang, Q. Zhang, X. Yan, L. Chai, Resveratrol inhibits monocrotaline-induced pulmonary arterial remodeling by suppression of SphK1-mediated NF-κB activation, Life sciences 210 (2018) 140-149.
[26] Z. Wang, Y. Chen, N. Labinskyy, T.-c. Hsieh, Z. Ungvari, J.M. Wu, Regulation of proliferation and gene expression in cultured human aortic smooth muscle cells by resveratrol and standardized grape extracts, Biochemical and biophysical research communications 346(1) (2006) 367-376.
[27] R.E. Morty, B. Nejman, G. Kwapiszewska, M. Hecker, A. Zakrzewicz, F.M. Kouri, D.M. Peters, R. Dumitrascu, W. Seeger, P. Knaus, Dysregulated bone morphogenetic protein signaling in monocrotaline-induced pulmonary arterial hypertension, Arteriosclerosis, thrombosis, and vascular biology 27(5) (2007) 1072-1078.
[28] J.Q. Liu, I.N. Zelko, E.M. Erbynn, J.S. Sham, R.J. Folz, Hypoxic pulmonary hypertension: role of superoxide and NADPH oxidase (gp91phox), American Journal of Physiology-Lung Cellular and Molecular Physiology 290(1) (2006) L2-L10.
[29] A. Sturrock, B. Cahill, K. Norman, T.P. Huecksteadt, K. Hill, K. Sanders, S. Karwande, J.C. Stringham, D.A. Bull, M. Gleich, Transforming growth factor-β1 induces Nox4 NAD (P) H oxidase and reactive oxygen species-dependent proliferation in human pulmonary artery smooth muscle cells, American Journal of Physiology-Lung Cellular and Molecular Physiology 290(4) (2006) L661-L673.
[30] C. McEvoy, M. de Gaetano, H.E. Giffney, B. Bahar, E.P. Cummins, E.P. Brennan, M. Barry, O. Belton, C.G. Godson, E.P. Murphy, NR4A receptors differentially regulate NF-κB signaling in myeloid cells, Frontiers in immunology 8 (2017) 7.
[31] R. Rodríguez-Calvo, A. Guadall, O. Calvayrac, M.A. Navarro, J. Alonso, B. Ferrán, A. de Diego, P. Muniesa, J. Osada, C. Rodríguez, Over-expression of neuron-derived orphan receptor-1 (NOR-1)

exacerbates neointimal hyperplasia after vascular injury, Human Molecular Genetics 22(10) (2013) 1949-1959.
[32] O. Boucherat, F. Potus, S. Bonnet, microRNA and pulmonary hypertension, microRNA: Medical Evidence, Springer2015, pp. 237-252.
[33] X.-G. Zhao, J.-Y. Hu, J. Tang, W. Yi, M.-Y. Zhang, R. Deng, S.-J. Mai, N.-Q. Weng, R.-Q. Wang, J. Liu, miR-665 expression predicts poor survival and promotes tumor metastasis by targeting NR4A3 in breast cancer, Cell death & disease 10(7) (2019) 1-21.
[34] Y.-y. Liu, W.-y. Zhang, C.-g. Wang, J.-a. Huang, J.-h. Jiang, D.-x. Zeng, Resveratrol prevented experimental pulmonary vascular remodeling via miR-638 regulating NR4A3/cyclin D1 pathway, Microvascular Research 130 (2020) 103988.
[35] Y. Ren, Y. Chen, X. Liang, Y. Lu, W. Pan, M. Yang, MiRNA-638 promotes autophagy and malignant phenotypes of cancer cells via directly suppressing DACT3, Cancer letters 390 (2017) 126-136.
[36] M. He, Y. Lin, Y. Tang, Y. Liu, W. Zhou, C. Li, G. Sun, M. Guo, miR-638 suppresses DNA damage repair by targeting SMC1A expression in terminally differentiated cells, Aging (Albany NY) 8(7) (2016) 1442.
[37] J. Chen, I.F. López-Moyado, H. Seo, C.-W.J. Lio, L.J. Hempleman, T. Sekiya, A. Yoshimura, J.P. Scott-Browne, A. Rao, NR4A transcription factors limit CAR T cell function in solid tumours, Nature 567(7749) (2019) 530-534.
[38] A.M. Ramirez-Herrick, S.E. Mullican, A.M. Sheehan, O.M. Conneely, Reduced NR4A gene dosage leads to mixed myelodysplastic/myeloproliferative neoplasms in mice, Blood 117(9) (2011) 2681-2690.
[39] H. Nakayama, T. Yaguchi, S. Yoshiya, T. Nishizaki, Resveratrol induces apoptosis MH7A human rheumatoid arthritis synovial cells in a sirtuin 1-dependent manner, Rheumatology international 32(1) (2012) 151-157.
[40] W. San Cheang, W.T. Wong, L. Wang, C.K. Cheng, C.W. Lau, R.C.W. Ma, A. Xu, N. Wang, Y. Huang, X.Y. Tian, Resveratrol ameliorates endothelial dysfunction in diabetic and obese mice through sirtuin 1 and peroxisome proliferator-activated receptor δ, Pharmacological research 139 (2019) 384-394.
[41] J.N. Feige, J. Auwerx, Transcriptional targets of sirtuins in the coordination of mammalian physiology, Current opinion in cell biology 20(3) (2008) 303-309.
[42] L. Li, H.-N. Zhang, H.-Z. Chen, P. Gao, L.-H. Zhu, H.-L. Li, X. Lv, Q.-J. Zhang, R. Zhang, Z. Wang, SIRT1 acts as a modulator of neointima formation following vascular injury in mice, Circulation research 108(10) (2011) 1180-1189.
[43] S. Yao, L.Y.-N. Fan, E.W.-F. Lam, The FOXO3-FOXM1 axis: A key cancer drug target and a modulator of cancer drug resistance, Seminars in cancer biology, Elsevier, 2018, pp. 77-89.
[44] A. Bourgeois, C. Lambert, K. Habbout, B. Ranchoux, S. Paquet-Marceau, I. Trinh, S. Breuils-Bonnet, R. Paradis, V. Nadeau, R. Paulin, FOXM1 promotes pulmonary artery smooth muscle cell expansion in pulmonary arterial hypertension, Journal of Molecular Medicine 96(2) (2018) 223-235.
[45] K. Zainabadi, C.J. Liu, L. Guarente, SIRT1 is a positive regulator of the master osteoblast transcription factor, RUNX2, PloS one 12(5) (2017) e0178520.
[46] G. Ruffenach, S. Chabot, V.F. Tanguay, A. Courboulin, O. Boucherat, F. Potus, J. Meloche, A. Pflieger, S. Breuils-Bonnet, V. Nadeau, Role for runt-related transcription factor 2 in proliferative and calcified vascular lesions in pulmonary arterial hypertension, American journal of respiratory and critical care medicine 194(10) (2016) 1273-1285.
[47] M. Shakibaei, P. Shayan, F. Busch, C. Aldinger, C. Buhrmann, C. Lueders, A. Mobasheri, Resveratrol mediated modulation of Sirt-1/Runx2 promotes osteogenic differentiation of mesenchymal stem cells: potential role of Runx2 deacetylation, PloS one 7(4) (2012) e35712.
[48] B. Ranchoux, J. Meloche, R. Paulin, O. Boucherat, S. Provencher, S. Bonnet, DNA damage and pulmonary hypertension, International Journal of Molecular Sciences 17(6) (2016) 990.
[49] M. Yanez, M. Jhanji, K. Murphy, R.M. Gower, M. Sajish, E. Jabbarzadeh, Nicotinamide augments the anti-inflammatory properties of resveratrol through PARP1 activation, Scientific reports 9(1) (2019) 1-10.

[50] S. Zhou, M.-T. Li, Y.-Y. Jia, J.-J. Liu, Q. Wang, Z. Tian, Y.-T. Liu, H.-Z. Chen, D.-P. Liu, X.-F. Zeng, Regulation of cell cycle regulators by SIRT1 contributes to resveratrol-mediated prevention of pulmonary arterial hypertension, BioMed research international 2015 (2015).
[51] S. Kanno, Y.-J.L. Wu, P.C. Lee, T.R. Billiar, C. Ho, Angiotensin-converting enzyme inhibitor preserves p21 and endothelial nitric oxide synthase expression in monocrotaline-induced pulmonary arterial hypertension in rats, Circulation 104(8) (2001) 945-950.
[52] D.W. Stacey, Cyclin D1 serves as a cell cycle regulatory switch in actively proliferating cells, Current opinion in cell biology 15(2) (2003) 158-163.
[53] L. Yu, Y. Tu, X. Jia, K. Fang, L. Liu, L. Wan, C. Xiang, Y. Wang, X. Sun, T. Liu, Resveratrol protects against pulmonary arterial hypertension in rats via activation of silent information regulator 1, Cellular Physiology and Biochemistry 42(1) (2017) 55-67.
[54] P. Shelton, A.K. Jaiswal, The transcription factor NF-E2-related factor 2 (Nrf2): a protooncogene?, The FASEB Journal 27(2) (2013) 414-423.
[55] B. Rodríguez-Iturbe, Y. Quiroz, M. Nava, L. Bonet, M. Chávez, J. Herrera-Acosta, R.J. Johnson, H.A. Pons, Reduction of renal immune cell infiltration results in blood pressure control in genetically hypertensive rats, American Journal of Physiology-Renal Physiology 282(2) (2002) F191-F201.
[56] A.A. Javkhedkar, Y. Quiroz, B. Rodriguez-Iturbe, N.D. Vaziri, M.F. Lokhandwala, A.A. Banday, Resveratrol restored Nrf2 function, reduced renal inflammation, and mitigated hypertension in spontaneously hypertensive rats, American Journal of Physiology-Regulatory, Integrative and Comparative Physiology 308(10) (2015) R840-R846.
[57] L. George, M.F. Lokhandwala, M. Asghar, Exercise activates redox-sensitive transcription factors and restores renal D1 receptor function in old rats, American Journal of Physiology-Renal Physiology 297(5) (2009) F1174-F1180.
[58] D. Schreier, T. Hacker, G. Song, N. Chesler, The role of collagen synthesis in ventricular and vascular adaptation to hypoxic pulmonary hypertension, Journal of biomechanical engineering 135(2) (2013).
[59] D. Xu, Y. Li, B. Zhang, Y. Wang, Y. Liu, Y. Luo, W. Niu, M. Dong, M. Liu, H. Dong, Resveratrol alleviate hypoxic pulmonary hypertension via anti-inflammation and anti-oxidant pathways in rats, International journal of medical sciences 13(12) (2016) 942.
[60] A.F. Wright, M.-A. Ewart, K. Mair, M. Nilsen, Y. Dempsie, L. Loughlin, M.R. Maclean, Oestrogen receptor alpha in pulmonary hypertension, Cardiovascular research 106(2) (2015) 206-216.
[61] E.D. Austin, T. Lahm, J. West, S.P. Tofovic, A.K. Johansen, M.R. MacLean, A. Alzoubi, M. Oka, Gender, sex hormones and pulmonary hypertension, Pulmonary circulation 3(2) (2013) 294-314.
[62] C.M. Klinge, K.A. Blankenship, K.E. Risinger, S. Bhatnagar, E.L. Noisin, W.K. Sumanasekera, L. Zhao, D.M. Brey, R.S. Keynton, Resveratrol and estradiol rapidly activate MAPK signaling through estrogen receptors α and β in endothelial cells, Journal of Biological Chemistry 280(9) (2005) 7460-7468.
[63] C.M. Klinge, N.S. Wickramasinghe, M.M. Ivanova, S.M. Dougherty, Resveratrol stimulates nitric oxide production by increasing estrogen receptor α-Src-caveolin-1 interaction and phosphorylation in human umbilical vein endothelial cells, The FASEB Journal 22(7) (2008) 2185-2197.
[64] A.R. Khandelwal, V.Y. Hebert, T.R. Dugas, Essential role of ER-α-dependent NO production in resveratrol-mediated inhibition of restenosis, American Journal of Physiology-Heart and Circulatory Physiology 299(5) (2010) H1451-H1458.
[65] J.-L. Balligand, Reducing damage through Nrf-2, Oxford University Press, 2013.
[66] C. Harris, J.M. Hansen, N rf2-Mediated Resistance to Oxidant-Induced Redox Disruption in Embryos, Birth Defects Research Part B: Developmental and Reproductive Toxicology 95(3) (2012) 213-218.
[67] M. Niso-Santano, R.A. González-Polo, J.M. Bravo-San Pedro, R. Gómez-Sánchez, I. Lastres-Becker, M.A. Ortiz-Ortiz, G. Soler, J.M. Morán, A. Cuadrado, J.M. Fuentes, Activation of apoptosis signal-regulating kinase 1 is a key factor in paraquat-induced cell death: modulation by the Nrf2/Trx axis, Free Radical Biology and Medicine 48(10) (2010) 1370-1381.

[68] L.A. Shimoda, G.L. Semenza, HIF and the lung: role of hypoxia-inducible factors in pulmonary development and disease, American journal of respiratory and critical care medicine 183(2) (2011) 152-156.
[69] L.E. Huang, J. Gu, M. Schau, H.F. Bunn, Regulation of hypoxia-inducible factor 1α is mediated by an O2-dependent degradation domain via the ubiquitin-proteasome pathway, Proceedings of the National Academy of Sciences 95(14) (1998) 7987-7992.
[70] J. Du, R. Xu, Z. Hu, Y. Tian, Y. Zhu, L. Gu, L. Zhou, PI3K and ERK-induced Rac1 activation mediates hypoxia-induced HIF-1α expression in MCF-7 breast cancer cells, PloS one 6(9) (2011).
[71] S. Frede, C. Stockmann, P. Freitag, J. Fandrey, Bacterial lipopolysaccharide induces HIF-1 activation in human monocytes via p44/42 MAPK and NF-κB, Biochemical Journal 396(3) (2006) 517-527.
[72] S.-J. Lee, A. Smith, L. Guo, T.-P. Alastalo, M. Li, H. Sawada, X. Liu, Z.-H. Chen, E. Ifedigbo, Y. Jin, Autophagic protein LC3B confers resistance against hypoxia-induced pulmonary hypertension, American journal of respiratory and critical care medicine 183(5) (2011) 649-658.
[73] J. Wu, Z. Yu, D. Su, BMP4 protects rat pulmonary arterial smooth muscle cells from apoptosis by PI3K/AKT/Smad1/5/8 signaling, International journal of molecular sciences 15(8) (2014) 13738-13754.
[74] B. Yi, J. Cui, J.-n. Ning, G.-s. Wang, G.-s. Qian, K.-z. Lu, Over-expression of PKGIα inhibits hypoxia-induced proliferation, Akt activation, and phenotype modulation of human PASMCs: The role of phenotype modulation of PASMCs in pulmonary vascular remodeling, Gene 492(2) (2012) 354-360.
[75] Z. Guan, L. Shen, H. Liang, H. Yu, B. Hei, X. Meng, L. Yang, Resveratrol inhibits hypoxia-induced proliferation and migration of pulmonary artery vascular smooth muscle cells by inhibiting the phosphoinositide 3-kinase/protein kinase B signaling pathway, Molecular Medicine Reports 16(2) (2017) 1653-1660.
[76] E.A. Goncharova, A.J. Ammit, C. Irani, R.G. Carroll, A.J. Eszterhas, R.A. Panettieri, V.P. Krymskaya, PI3K is required for proliferation and migration of human pulmonary vascular smooth muscle cells, American Journal of Physiology-Lung Cellular and Molecular Physiology 283(2) (2002) L354-L363.
[77] O. RACHID, M. ALKHALAF, Resveratrol regulation of PI3K-AKT signaling pathway genes in MDA-MB-231 breast cancer cells, Cancer Genomics-Proteomics 3(6) (2006) 383-388.
[78] B. Chen, J. Xue, X. Meng, J.L. Slutzky, A.E. Calvert, L.G. Chicoine, Resveratrol prevents hypoxia-induced arginase II expression and proliferation of human pulmonary artery smooth muscle cells via Akt-dependent signaling, American Journal of Physiology-Lung Cellular and Molecular Physiology 307(4) (2014) L317-L325.
[79] R. Shivakrupa, A. Bernstein, N. Watring, D. Linnekin, Phosphatidylinositol 3'-kinase is required for growth of mast cells expressing the kit catalytic domain mutant, Cancer research 63(15) (2003) 4412-4419.
[80] M.C. Zimmerman, E. Lazartigues, R.V. Sharma, R.L. Davisson, Hypertension caused by angiotensin II infusion involves increased superoxide production in the central nervous system, Circulation research 95(2) (2004) 210-216.
[81] L. Tang, F. Dai, Y. Liu, X. Yu, C. Huang, Y. Wang, W. Yao, RhoA/ROCK signaling regulates smooth muscle phenotypic modulation and vascular remodeling via the JNK pathway and vimentin cytoskeleton, Pharmacological research 133 (2018) 201-212.
[82] C.N. Amaya, D.C. Mitchell, B.A. Bryan, Rho kinase proteins display aberrant upregulation in vascular tumors and contribute to vascular tumor growth, BMC cancer 17(1) (2017) 485.
[83] N.C. Lopez, G. Ebensperger, E.A. Herrera, R.V. Reyes, G. Calaf, G. Cabello, F.A. Moraga, F.A. Beñaldo, M. Diaz, J.T. Parer, Role of the RhoA/ROCK pathway in high-altitude associated neonatal pulmonary hypertension in lambs, American Journal of Physiology-Regulatory, Integrative and Comparative Physiology 310(11) (2016) R1053-R1063.

[84] Y. Fukumoto, A. Takaki, S. Tawara, J. Ohashi, M. Nakano, T. Tada, K. Saji, K. Sugimura, H. Fujita, Y. Hoshikawa, Evidence for Rho-kinase activation in patients with pulmonary arterial hypertension, Circulation Journal 73(9) (2009) 1731-1739.
[85] C. Guilluy, V. Sauzeau, M. Rolli-Derkinderen, P. Guérin, C. Sagan, P. Pacaud, G. Loirand, Inhibition of RhoA/Rho kinase pathway is involved in the beneficial effect of sildenafil on pulmonary hypertension, British journal of pharmacology 146(7) (2005) 1010-1018.
[86] A.D. Nalli, S. Rajagopal, S. Mahavadi, J.R. Grider, K.S. Murthy, Inhibition of RhoA-dependent pathway and contraction by endogenous hydrogen sulfide in rabbit gastric smooth muscle cells, American Journal of Physiology-Cell Physiology 308(6) (2015) C485-C495.
[87] N. Pannu, A. Bhatnagar, Resveratrol: From enhanced biosynthesis and bioavailability to multitargeting chronic diseases, Biomedicine & Pharmacotherapy 109 (2019) 2237-2251.
[88] M. Koushki, N. Amiri-Dashatan, N. Ahmadi, H.A. Abbaszadeh, M. Rezaei-Tavirani, Resveratrol: A miraculous natural compound for diseases treatment, Food Science & Nutrition 6(8) (2018) 2473-2490.
[89] L.D. Maston, D.T. Jones, W. Giermakowska, T.A. Howard, J.L. Cannon, W. Wang, Y. Wei, W. Xuan, T.C. Resta, L.V.G. Bosc, Central role of T helper 17 cells in chronic hypoxia-induced pulmonary hypertension, American Journal of Physiology-Lung Cellular and Molecular Physiology (2017).
[90] C. Li, G. Peng, J. Long, P. Xiao, X. Zeng, H. Yang, Protective effects of resveratrol and SR1001 on hypoxia-induced pulmonary hypertension in rats, Clinical and Experimental Hypertension 42(6) (2020) 519-526.
[91] P.M. Werchan, W.R. Summer, A.M. Gerdes, K. McDonough, Right ventricular performance after monocrotaline-induced pulmonary hypertension, American Journal of Physiology-Heart and Circulatory Physiology 256(5) (1989) H1328-H1336.
[92] D.L. Yang, H.G. Zhang, Y.L. Xu, Y.H. Gao, X.J. Yang, X.Q. Hao, X.H. Li, Resveratrol inhibits right ventricular hypertrophy induced by monocrotaline in rats, Clinical and Experimental Pharmacology and Physiology 37(2) (2010) 150-155.
[93] E. Vázquez-Garza, J. Bernal-Ramírez, C. Jerjes-Sánchez, O. Lozano, E. Acuña-Morín, M. Vanoye-Tamez, M.R. Ramos-González, H. Chapoy-Villanueva, L. Pérez-Plata, L. Sánchez-Trujillo, G. Torre-Amione, A. Ramírez-Rivera, G. García-Rivas, Resveratrol Prevents Right Ventricle Remodeling and Dysfunction in Monocrotaline-Induced Pulmonary Arterial Hypertension with a Limited Improvement in the Lung Vasculature, Oxidative Medicine and Cellular Longevity 2020 (2020).
[94] W.X. Xin, Q.L. Li, L. Fang, L.K. Zhong, X.W. Zheng, P. Huang, Preventive Effect and Mechanism of Ethyl Acetate Extract of Sceptridium ternatum in Monocrotaline-Induced Pulmonary Arterial Hypertension, Chinese Journal of Integrative Medicine 26(3) (2020) 205-211.
[95] D.N. Wilson, S.E. Schacht, L. Al-Nakkash, J.R. Babu, T.L. Broderick, Resveratrol prevents pulmonary trunk remodeling but not right ventricular hypertrophy in monocrotaline-induced pulmonary hypertension, Pathophysiology 23(4) (2016) 243-250.
[96] T.-M. Lu, J.-Y. Tsai, Y.-C. Chen, C.-Y. Huang, H.-L. Hsu, C.-F. Weng, C.-C. Shih, C.-P. Hsu, Downregulation of Sirt1 as aging change in advanced heart failure, Journal of biomedical science 21(1) (2014) 57.
[97] J. Maizel, S. Xavier, J. Chen, C.H.S. Lin, R. Vasko, M.S. Goligorsky, Sirtuin 1 ablation in endothelial cells is associated with impaired angiogenesis and diastolic dysfunction, American Journal of Physiology-Heart and Circulatory Physiology 307(12) (2014) H1691-H1704.
[98] E. Vázquez-Garza, J. Bernal-Ramírez, C. Jerjes-Sánchez, O. Lozano, E. Acuña-Morín, M. Vanoye-Tamez, M.R. Ramos-González, H. Chapoy-Villanueva, L. Pérez-Plata, L. Sánchez-Trujillo, Resveratrol Prevents Right Ventricle Remodeling and Dysfunction in Monocrotaline-Induced Pulmonary Arterial Hypertension with a Limited Improvement in the Lung Vasculature, Oxidative medicine and cellular longevity 2020 (2020).

[99] B. Olas, B. Wachowicz, J. Saluk-Juszczak, T. Zieliński, Effect of resveratrol, a natural polyphenolic compound, on platelet activation induced by endotoxin or thrombin, Thrombosis research 107(3-4) (2002) 141-145.
[100] E. Frankel, J. German, J. Kinsella, E. Parks, J. Kanner, Inhibition of oxidation of human low-density lipoprotein by phenolic substances in red wine, The Lancet 341(8843) (1993) 454-457.
[101] J.G. Zou, Y.Z. Huang, Q. Chen, E.H. Wei, T.c. Hsieh, J.M. Wu, Resveratrol inhibits copper ion-induced and azo compound-initiated oxidative modification of human low density lipoprotein, IUBMB Life 47(6) (1999) 1089-1096.
[102] T. Wallerath, G. Deckert, T. Ternes, H. Anderson, H. Li, K. Witte, U. Förstermann, Resveratrol, a polyphenolic phytoalexin present in red wine, enhances expression and activity of endothelial nitric oxide synthase, Circulation 106(13) (2002) 1652-1658.
[103] U.R. Pendurthi, L.V.M. Rao, Resveratrol suppresses agonist-induced monocyte adhesion to cultured human endothelial cells, Thrombosis research 106(4) (2002) 243-248.
[104] A. Di Santo, A. Mezzetti, E. Napoleone, R. Di Tommaso, M. Donati, G. De Gaetano, R. Lorenzet, Resveratrol and quercetin down-regulate tissue factor expression by human stimulated vascular cells, Journal of Thrombosis and Haemostasis 1(5) (2003) 1089-1095.
[105] M.A. Carluccio, L. Siculella, M.A. Ancora, M. Massaro, E. Scoditti, C. Storelli, F. Visioli, A. Distante, R. De Caterina, Olive oil and red wine antioxidant polyphenols inhibit endothelial activation: antiatherogenic properties of Mediterranean diet phytochemicals, Arteriosclerosis, thrombosis, and vascular biology 23(4) (2003) 622-629.
[106] M.J. Eagleton, P.K. Henke, C.E. Luke, A.E. Hawley, A. Bedi, B.S. Knipp, T.W. Wakefield, L.J. Greenfield, Inflammation and intimal hyperplasia associated with experimental pulmonary embolism, Journal of vascular surgery 36(3) (2002) 581-588.
[107] F. Langer, R. Schramm, M. Bauer, D. Tscholl, T. Kunihara, H.-J. Scha, Cytokine response to pulmonary thromboendarterectomy, Chest 126(1) (2004) 135-141.
[108] J.E. Rectenwald, K.B. Deatrick, P. Sukheepod, E.M. Lynch, A.J. Moore, D.M. Moaveni, N.A. Deywer, C.E. Luke, G.R. Upchurch Jr, T.W. Wakefield, Experimental pulmonary embolism: effects of the thrombus and attenuation of pulmonary artery injury by low-molecular-weight heparin, Journal of vascular surgery 43(4) (2006) 800-808.
[109] S.R. Green, K.H. Han, Y. Chen, F. Almazan, I.F. Charo, Y.I. Miller, O. Quehenberger, The CC chemokine MCP-1 stimulates surface expression of CX3CR1 and enhances the adhesion of monocytes to fractalkine/CX3CL1 via p38 MAPK, The Journal of Immunology 176(12) (2006) 7412-7420.
[110] C. Chen, J.W. Lin, G.P. Li, Z.C. Ren, Y.Q. Li, J.P. Yan, L.X. Wang, Resveratrol down-regulates acute pulmonary thromboembolism-induced pulmonary artery hypertension and monocyte chemoattractant protein-1 in rats, Chinese Pharmacological Bulletin 33(10) (2017) 1436-1441.
[111] Z. Sun, W. Zheng, J. Teng, Z. Fang, C. Lin, Resveratrol Reduces Kidney Injury in a Rat Model of Uremia and is Associated with Increased Expression of Heat Shock Protein 70 (Hsp70), Medical Science Monitor: International Medical Journal of Experimental and Clinical Research 26 (2020) e919086-1.
[112] L. Liu, C. Fu, M. Yan, H. Xie, S. Li, Q. Yu, S. He, J. He, Resveratrol modulates intestinal morphology and HSP70/90, NF-κB and EGF expression in the jejunal mucosa of black-boned chickens on exposure to circular heat stress, Food & function 7(3) (2016) 1329-1338.
[113] P.K. Chakraborty, S.B. Mustafi, S. Ganguly, M. Chatterjee, S. Raha, Resveratrol induces apoptosis in K562 (chronic myelogenous leukemia) cells by targeting a key survival protein, heat shock protein 70, Cancer science 99(6) (2008) 1109-1116.
[114] I.M. Kapetanovic, M. Muzzio, Z. Huang, T.N. Thompson, D.L. McCormick, Pharmacokinetics, oral bioavailability, and metabolic profile of resveratrol and its dimethylether analog, pterostilbene, in rats, Cancer chemotherapy and pharmacology 68(3) (2011) 593-601.

[115] F.S. Aldawsari, C.A. Velázquez-Martínez, 3, 4', 5-trans-Trimethoxystilbene; a natural analogue of resveratrol with enhanced anticancer potency, Investigational New Drugs 33(3) (2015) 775-786.
[116] G. Gao, X. Wang, X. Qin, X. Jiang, D. Xiang, L. Xie, J. Hu, J. Gao, Effects of trimethoxystilbene on proliferation and apoptosis of pulmonary artery smooth muscle cells, Cell biochemistry and biophysics 64(2) (2012) 101-106.
[117] J. Shu, W. Liu, F. Han, F. Luo, Trimethoxystilbene Reduces Nuclear Factor Kappa B, Interleukin-6, and Tumor Necrosis Factor-α Levels in Rats with Pulmonary Artery Hypertension, BioMed Research International 2019 (2019).
[118] D.M. Tabima, S. Frizzell, M.T. Gladwin, Reactive oxygen and nitrogen species in pulmonary hypertension, Free Radical Biology and Medicine 52(9) (2012) 1970-1986.
[119] Y.-S. Zhang, L. He, B. Liu, N.-S. Li, X.-J. Luo, C.-P. Hu, Q.-L. Ma, G.-G. Zhang, Y.-J. Li, J. Peng, A novel pathway of NADPH oxidase/vascular peroxidase 1 in mediating oxidative injury following ischemia–reperfusion, Basic research in cardiology 107(3) (2012) 266.
[120] T.-T. Li, Y.-S. Zhang, L. He, B. Liu, R.-Z. Shi, G.-G. Zhang, J. Peng, Inhibition of vascular peroxidase alleviates cardiac dysfunction and apoptosis induced by ischemia–reperfusion, Canadian journal of physiology and pharmacology 90(7) (2012) 851-862.
[121] S. Tasneem, B. Liu, B. Li, M.I. Choudhary, W. Wang, Molecular pharmacology of inflammation: Medicinal plants as anti-inflammatory agents, Pharmacological research 139 (2019) 126-140.
[122] B. Liu, X.-J. Luo, Z.-B. Yang, J.-J. Zhang, T.-B. Li, X.-J. Zhang, Q.-L. Ma, G.-G. Zhang, C.-P. Hu, J. Peng, Inhibition of NOX/VPO1 pathway and inflammatory reaction by trimethoxystilbene in prevention of cardiovascular remodeling in hypoxia-induced pulmonary hypertensive rats, Journal of cardiovascular pharmacology 63(6) (2014) 567-576.
[123] M. Smola, T. Vandamme, A. Sokolowski, Nanocarriers as pulmonary drug delivery systems to treat and to diagnose respiratory and non respiratory diseases, International journal of nanomedicine 3(1) (2008) 1.
[124] M.C. Fontana, T.L. Durli, A.R. Pohlmann, S.S. Guterres, R.C.R. Beck, Polymeric controlled release inhalable powder produced by vibrational spray-drying: one-step preparation and in vitro lung deposition, Powder technology 258 (2014) 49-59.
[125] K. Schmid, C. Arpagaus, W. Friess, Evaluation of the Nano Spray Dryer B-90 for pharmaceutical applications, Pharmaceutical development and technology 16(4) (2011) 287-294.
[126] F.A. Dimer, M. Ortiz, A.R. Pohlmann, S.S. Guterres, Inhalable resveratrol microparticles produced by vibrational atomization spray drying for treating pulmonary arterial hypertension, Journal of Drug Delivery Science and Technology 29 (2015) 152-158.